

Amendment to the Specification:

Please amend the specification as follows:

Please replace the pending title with the following new title:

PHYTASE EXPRESSION SYSTEMS AND METHODS OF MAKING AND USING THEM

Please replace the paragraph of lines 13 to 21 on page 6 with the following amended paragraph:

2.1.2--Nutritional Concerns: Many potentially nutritious plants, including particularly their seeds, contain appreciable amounts of nutrients, e.g. phosphate, that are associated with phytate in a manner such that these nutrients are not freely available upon consumption. The unavailability of these nutrients is overcome by some organisms, including cows and other ruminants, that have a sufficient digestive ability--largely derived from the presence of symbiotic life forms in their digestive tracts--to hydrolyze phytate and liberate the associated nutrients. However, the majority of species of farmed animals, including pigs, fish, chickens, turkeys, as well as other non-ruminant organisms including man, are unable to efficiently liberate these nutrients after ingestion [[injection]].

Please replace the paragraph of lines 22 to 25 on page 6 with the following amended paragraph:

Consequently, phytate-containing foodstuffs require supplementation with exogenous nutrients and/or with a source of phytase activity in order to [[amend]] amend their deficient nutritional offerings upon consumption by a very large number of species of organisms.

Please replace the paragraph spanning pages 6 and 7 with the following amended paragraph:

2.1.3--Ex vivo Processing Concerns: In yet another aspect, the presence of unhydrolyzed phytate leads to problematic consequences in ex vivo processes including--but not limited to--the processing of foodstuffs. In but merely one exemplification, as described in EP0321004-B1 (Vaara et al), there is a step in the processing of corn and sorghum kernels whereby the hard kernels are steeped in water to soften them. Water-soluble [[substances]]

substances that leach out during this process become part of a corn steep liquor, which is concentrated by evaporation. Unhydrolyzed phytic acid in the corn steep liquor, largely in the form of calcium and magnesium salts, is associated with phosphorus and deposits an undesirable sludge with proteins and metal ions. This sludge is problematic in the evaporation, transportation and storage of the corn steep liquor. Accordingly, the instantly disclosed phytase molecules--either alone or in combination with other reagents (including but not limited to enzymes, including proteases)--are serviceable not only in this application (e.g., for prevention of the unwanted [[slugde]] sludge) but also in other applications where phytate hydrolysis is desirable.

Please replace the paragraph of lines 1 to 5 on page 9 with the following amended paragraph:

Consequently, there is a need for means to achieve efficient and cost effective hydrolysis of phytate in various applications. Particularly, there is a need for means to optimize the [[hyrolysis]] hydrolysis of phytate in commercial applications. In a particular aspect, there is a need to optimize commercial treatment methods that improve the nutritional offerings of phytate-containing foodstuffs for consumption by humans and farmed animals.

Please replace the paragraph of lines 10 to 21 on page 8 with the following amended paragraph:

2.1.5--Environmental Concerns: An environmental consequence is that the consumption of phytate-containing foodstuffs by any organism species that is phytase-deficient--regardless of whether the foodstuffs are supplemented with minerals--leads to fecal pollution resulting from the excretion of unabsorbed minerals. This pollution has a negative impact not only on the immediate habitat but consequently also on the surrounding waters. The environmental alterations occur primarily at the bottom of the food chain, and therefore have the potential to permeate upwards and throughout an ecosystem to effect permanent and catastrophic damage--particularly after years of continual pollution. This problem has the potential to manifest itself in any area where concentrated phytate processing occurs--including in vivo (e.g. by animals in areas of livestock production, zoological grounds, wildlife refuges, etc.) and in

vitro (e.g. in commercial corn wet milling, ~~[[ceral]]~~ cereal steeping processes, etc.) processing steps.

Please delete the paragraph of lines 21 to 23 on page 15.

Please replace the paragraph of lines 4 to 6 on page 18 with the following amended paragraph:

Figure 1A, Figure 1B, and Figure 1C show ~~[[FIG. 1 shows]]~~ the nucleotide (SEQ ID NO:1) and deduced amino acid (SEQ ID NO:2) sequences the enzyme of the present invention. Sequencing was performed using a 378 automated DNA sequencer (Applied Biosystems, Inc.).

Please delete the paragraph of lines 15 to 21 on page 31.

Please replace the paragraph of lines 3 to 8 on page 52 with the following amended paragraph:

6.3.2--Administration to organisms: In a non-limiting ~~[[apsect]]~~ aspect, the recombinant phytase can be consumed by organisms and retains activity upon consumption. In another exemplification, transgenic ~~[[approches]]~~ approaches can be used to achieve expression ~~[[fo]]~~ of the recombinant phytase--preferably in a controlled fashion (methods are available for controlling expression of transgenic molecules in time-specific and tissue specific manners).

Please replace the abstract (paragraph of lines 1 to 5) on page 87 with the following amended paragraph:

The invention provides a ~~[[A]]~~ purified or recombinant phytase enzyme (SEQ ID NO:2) initially derived from Escherichia coli B. The enzyme has a molecular weight of about 47.1 kilodaltons and has phytase activity (SEQ ID NO:2). The enzyme can be produced from native or recombinant host cells and can be used to aid in the digestion of phytate where desired. In particular, the phytase of the present invention can be used in foodstuffs to improve the feeding value of phytate rich ingredients.